Results of Testing

Chemical Name	CAS No.	Study Code/Type	Protocol/Guideline	Species	Exposure	Dose/Concentration	No. per Group	Results	Reference
Benzidine, *o*-toluidine, *o*-dianisidine	Not available	EFBDEG Aerobic biodegradation	Non-TSCA Protocol/ Guideline (docket OPTS-42001)	Not relevant	aerobic, conditions similar to OECD Ready Biodegrad-ability Test 301A in regard to inoculum level and test medium.	3 mg/l	Not relevant	The chemicals studies, a selection of aromatic amines, possible biodegradation products of azo dyes, including *o*-dianisidine and 3,3'-dichlorobenzidine. Under the test conditions these products were not "readily biodegradable" but their "inherent biodegradability" was demonstrated. Results were confirment using the OECD Inherent Biodegradability Test 302 B (Zahn-Wellens test).	ETAD (The Ecological & Toxicol. Assoc. of the Dyestuffs Mfg. Industry) Project E 3011, OTS0507287
Benzidine, *o*-toluidine, *o*-dianisidine	Not available	EFBDEG Aerobic biodegradation	Non-TSCA Protocol/ Guideline (docket OPTS-42001)	Not relevant	aerobic conditions, 28 days, sewage inoculum	20 mg/l DOC	Not relevant	Results indicate the "readily biodegradable" of te 4 aromatic amines (aniline, p-anisidine, p-phenetidine and o-toluidine) and the "inherent biodegradability" of o-dianisidine and 3,3'-dichlorobenzidine. Therefore, if azo dyes are anaerobically cleaved to these amines, it is unlikely that they will remain unchanged in the environment.	Brown, D., et al. The aerobic biodegrad-ability of primary aromatic amines, ETAD, Docket OPTS-42002
Benzidine, *o*-toluidine, *o*-dianisidine	Not available	EFBDEG Anaerobic biodegradation	Non-TSCA Protocol/ Guideline (docket OPTS-42001)	Not relevant	anaerobic conditions	Not reported	Not relevant	Under the test conditions a moderate rate of primary degradation was observed with Direct Red 7, Acid Red 114, and Direct Blue 15.	ETAD Project E 3010, OTS0507287
Benzidine, *o*-toluidine, *o*-dianisidine	Not available	EFBDEG Anaerobic biodegradation	Non-TSCA Protocol/ Guideline (docket OPTS-42001)	Not relevant	35 °C, anaerobic conditions 42 days	Not reported	Not relevant	Studies were performed on 22 dyes chosed to be representative of major classes of dyestuffs and included Direct Red 7 as a positive control. The results show that with the single exception of Acid Blue 80 all the dyestuffs tested can show a substantial degree of colour removal and thus it seems that the breakdown of dyestuffs in the environment is likely to be initiated under anaerobic conditions.	Brown, D., et al. The degradation of dye-stuffs: Part 1: Primary biodegrad-ation under anaerobic conditions, ETAD, Docket OPTS-42002
Benzidine, *o*-toluidine, *o*-dianisidine	Not available	HEADME Pharmacokinetics	Non-TSCA Protocol/ Guideline (docket OPTS-42001)	rats	Not reported	Not reported	Not reported	Experiments were performed on C-14-labeled Direct Blue 15 and Direct Red 2. The minimum detectable levels of both dyes in feces were 0.2 ppm. Based on radioassays, 74% of each dose was excreted via the feces; however, HPLC assays showed that only 11% of each dose was present as intact dye in the excrement.	Levine, R.A., et al. 1982. J. Anal Toxicol 6: July/ August., FDA and NCTR (Natl Ctr Toxicol Research) OTS0507293

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Benzidine, *o*-toluidine, *o*-dianisidine	Not available	HEADME Pharmacokinetics	Non-TSCA Protocol/ Guideline (docket OPTS-42001)	rats	oral (single dose)	12 mg/kg	Not reported	The metabolism of Direct Blue 15 and Direct Red 2 in rats was studied. The base (DiMxBzd) of Direct Blue 15 was more extensively metabolized and most of the 14C in various extracts were identified as known metabolites. The base (DiMeBzd) of Direct Red 2 was more extensively metabolized with a small percentage of 14C identified as know metabolites. Distribution studies showed that liver, kidney, and lung accumulated and retained higher levels of 14C than other tissues (at 72 hrs). Peak levels of 14C, which occurred 8-12 hours after dosing were significantly higher with Direct Red 2 than Direct Blue 15.	Bowman, M.C., et al. 1982. J. Anal. Toxicol. 6: July/ August., NIOSH, FDA, and NCTR OTS0507294
Benzidine, *o*-toluidine, *o*-dianisidine	Not available	HEADME Pharmacokinetics	Non-TSCA Protocol/ Guideline (docket OPTS-42001)	rats	Not reported	2 mg	Not reported	Peak levels of metabolites were excreted either 0-12 or 12-24 hr after the dyes were administered and, in seven of nine instances, no metabolites persisted in the urine after 48 hr. Minimum detectable levels of all metabolites were 12 ppb or less. All nine dyes were shown to be converted to measurable levels of their benzidine-congener-based metabolites in rats.	OTS0507292, NTP (National Toxicology Program) and NCTR
Benzidine, *o*-toluidine, *o*-dianisidine	Not available	HEADME Pharmacokinetics	Non-TSCA Protocol/ Guideline (docket OPTS-42001)	rats	Not reported	2 mg	Not reported	Nine azo dyes based on dimethyl-, dimethoxy-, or dichlorobenzidene were studied to to determine whether free amine cogeners, their metabolites or conjugates were excreted in the urine. All 9 dyes were converted to measureable levels of their benzidine-cogener-based metabolites. Peak levels of metabolites were excreted either 0-12 or 12-24 hr after the dyes were administered and, in seven of nine instances, no metabolites persisted in the urine after 48 hr. Minimum detectable levels of all metabolites were 12 ppb or less.	OTS0507292, NTP and NCTR
Benzidine, *o*-toluidine, *o*-dianisidine	Not available	HECTOXTRFM Cell transformation	Non-TSCA Protocol/ Guideline (docket OPTS-42001)	hamster (kidney BHK21 cells)	*in vitro*	Not reported	Not reported	Direct Blue 14 and Direct Blue 53 produced positive results.	ETAD Project T 2002, OTS0507287
Benzidine, *o*-toluidine, *o*-dianisidine	Not available	HEGTOXMUTA *Salmonella* microsome mutation test	Non-TSCA Protocol/ Guideline (docket OPTS-42001)	*Salmonella typhimurium*	*in vitro*, with and without S-9 activation	Not reported	Not reported	Direct Blue 14 produced negative results. Direct Blue 53 produced a positive result with activation.	ETAD Project T 2002, OTS0507287

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Benzidine, *o*-toluidine, *o*-dianisidine	Not available	HEGTOXMUTA *Salmonella* microsome mutation test	Non-TSCA Protocol/ Guideline (docket OPTS-42001)	*Salmonella typhimurium*	*in vitro*, with and without S-9 activation	20, 100, 500, 2500, and 5000 µg/plate	Not reported	A dose-dependent mutagenic effect was observed for Acid Red 114 with the addition of S-9 mix with a maximal increase in the mutation rate by a factor of 4 from 250 µg to 1000 µg using TA 98 and from 500 µg to 1000 µg using TA 1538. A decrease in the number of colonies was observed from a dose greater than 1000 µg/plate. Inconclusive results were obtained using the tester strain TA 100.	ETAD Project T 2015-3, Docket OPTS-42002
Benzidine, *o*-toluidine, *o*-dianisidine	Not available	HESTOX Subacute toxicity	Non-TSCA Protocol/ Guideline (docket OPTS-42001)	rats	oral (gavage), 22 doses over 30 days	1000 mg/kg b.w.	Not reported	All products, including Direct Blue 15,were tolerated without irreversible signs of toxicity and exhibited very low cumulative toxicity.	ETAD Project T 2014, OTS0507287, Leist, K.H.. Ecotox & Environ Safety. 1982. 6: 457-463.